

A multivariate approach to the identification of unionid glochidia with emphasis on Species at Risk in Southern Ontario

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ABSTRACT

Tremblay, M.E.M., Morris, T.J., Ackerman, J.D. 2015. A multivariate approach to the identification of unionid glochidia with emphasis on Species at Risk in Southern Ontario. Can. Manuscr. Rep. Fish. Aquat. Sci. 3057: vii + 51 p.

Freshwater mussels in the family Unionidae have experienced severe population declines as a result of multiple factors, including their complex life history and associated dependence on fish hosts to complete their life cycle. Understanding this life history -- in particular, identification of host fish species and the timing of glochidia release -- is crucial to mussel conservation. One way to accomplish this task is to employ morphometrics (i.e., glochidia shape and size; specifically, height, hinge length, and length) to identify the glochidia present on the gills of fish or in the water column. Using Discriminant Function Analysis, four river-specific (Ausable River, Grand River, Sydenham River, and Thames River) models and one regional (Southern Ontario) model were created. All models were able to distinguish between mussel species based on their glochidial dimensions (statistically significant, $p < 0.05$) with classification success ranging between 71.1% and 77.9%. The classification success of a given species varied based on the distinctiveness of the shape and size of its glochidia. This report describes a convenient and cost-effective method for the identification of unknown glochidia in Southern Ontario, which may improve understanding and consequently, the ability to conserve freshwater unionid mussel populations.

RÉSUMÉ

Tremblay, M.E.M., Morris, T.J., Ackerman, J.D. 2015. A multivariate approach to the identification of unionid glochidia with emphasis on Species at Risk in Southern Ontario. Can. Manuscr. Rep. Fish. Aquat. Sci. 3057: vii + 51 p.

Les moules d'eau douce de la famille des unionidés ont connu un déclin important de leurs populations en raison de facteurs multiples, notamment leur cycle biologique complexe et leur dépendance à des poissons-hôtes pour compléter ce cycle. L'acquisition d'une meilleure connaissance de ce cycle biologique, plus précisément l'identification des espèces de poissons-hôtes et le moment de la libération des glochidies, est cruciale pour la conservation des moules. Une des façons d'accomplir cette tâche est d'utiliser les données morphométriques (c.-à-d. les données sur la taille et la forme des glochidies; plus particulièrement la hauteur, la longueur des charnières et la longueur) afin d'identifier les glochidies présentes sur les branchies des poissons ou dans la colonne d'eau. À l'aide de l'analyse discriminante des données, des modèles ont été créés : un modèle pour chacune des quatre rivières (rivière Ausable, rivière Grand, rivière Sydenham et rivière Thames) et un modèle régional (sud-ouest de l'Ontario). Tous les modèles ont permis de différencier les espèces de moules d'après les dimensions de

leur glochidies (statistiquement significatif = $p < 0,05$) avec un taux de classification réussie de l'ordre de 71,1 à 77,9 %. Le succès de la classification d'une espèce de moules donnée variait en fonction du caractère distinctif de la forme et de la taille de ses glochidies. Le présent rapport décrit une méthode pratique et économique pour identifier les glochidies inconnues du sud-ouest de l'Ontario. Cette méthode est susceptible d'améliorer les connaissances sur l'espèce et, par le fait même, la capacité de conserver les populations de moules d'eau douce de la famille des unionidés.

1.0 INTRODUCTION

Ontario's lower Great Lakes region is home to Canada's most diverse freshwater mussel (Unionidae) fauna. Forty-one of the 55 species that occur in Canada are found in this region (Metcalf-Smith et al. 2005) and over half of these (21 species) are found nowhere else in Canada (Clarke 1981, as cited by Metcalf-Smith et al. 2000). Many of these species have experienced declines due to a variety of factors, including invasive species (in particular, dreissenid mussels), habitat loss and degradation, and reductions in water quality (primarily as a result of urbanization and agriculture; Morris and Burridge, 2006; Fisheries and Oceans Canada, 2013). As a result, over one third of the region's mussel species have been assessed as at risk by the Committee on the Status of Endangered Wildlife in Canada (COSEWIC) (www.COSEWIC.gc.ca, accessed October 2013).

Due to the complex, obligate parasitic reproductive strategy employed by species within the Family Unionidae, the identification of the host fishes of these Threatened and Endangered mussels is a main objective of Recovery Strategies created under the *Species at Risk Act* (e.g., Morris and Burridge, 2006; Fisheries and Oceans Canada, 2013). For a fish species to act as a host for a mussel there are three necessary phases: 1) encounter; 2) infestation; and 3) metamorphosis (Schwalb et al. 2010; McNichols et al. 2011). During the encounter phase, viable mussel glochidia must come in direct contact with an individual from a suitable host species. The infestation phase requires the successful attachment of the glochidia to the host and this is then followed by the metamorphosis of the glochidia into viable juveniles (Barnhart et al. 2008). A primary experimental means of identifying potential host fishes is through the execution of laboratory host infestation experiments. During these experiments, potential host fishes are exposed to glochidia of mussel and both infestation and metamorphosis can be measured directly (e.g., McNichols 2007; McNichols et al. 2011). While laboratory experiments permit the evaluation of the infestation and metamorphosis phases, they provide no information about the encounter phase. In nature, the encounter phase is regulated by a variety of characteristics of both the mussel and the potential hosts, including distribution and habitat preferences (i.e., spatial overlap; McNichols et al. 2011) and life history characters (i.e., temporal overlap). In order to determine whether fish species that facilitate the laboratory metamorphosis of unionid mussels become infested and serve as hosts in nature, it is necessary to investigate all aspects of the encounter phase (timing, abundance, and diversity of unionid glochidial availability, both in the water column and on potential wild-caught hosts; Neves and Widlak 1988, Haag and Warren 2003).

In order to achieve this objective of investigating the encounter phase, the ability to classify the glochidia taxonomically (ideally to species-level) is necessary. There are currently two principal ways of doing so: first, through genetic methods (e.g., barcoding) and second, through morphometrics (i.e., analysis of glochidia size and shape). Both of these methodologies can and have been used to answer important questions about unionid ecology (e.g., Neves and Widlak 1988; Kennedy and Haag 2005; Kneeland and Rhymer 2008; Boyer et

al. 2011; Culp et al. 2011). However, morphometric models can be made widely available and permit the rapid and inexpensive identification of glochidia.

The objective of the work described here is to develop a methodology (specifically, a series of discriminant models) that can be used across southern Ontario to quickly identify glochidia of unknown species using their shell dimensions. The application of these models in the future will permit a greater understanding of the complex reproductive biology and life history of these SAR, including assisting with the identification of host fishes and the determination of the timing of glochidial release and infestation periods.

2.0 METHODS

A list of mussel species was compiled for each river of interest (Ausable, Grand, Sydenham, and Thames rivers), as well as an overall list of mussel species for southern Ontario using relevant literature (Metcalf et al. 2000; Metcalf et al. 2007; McNichols 2007; McNichols-O'Rourke et al. 2012; Fisheries and Oceans Canada (DFO) Lower Great Lakes Unionid database). Models included all freshwater mussel species that occur presently or historically in the specified waterbody or region.

Based on this, 27 unionid species were identified as occurring in the Ausable River, 32 species in the Grand River, 35 in the Sydenham River, and 35 in the Thames River (Table 1). Combining unique species across all watersheds of southern Ontario resulted in a potential pool of 36 species.

In order to build the models, raw data for length, hinge length, and height (Figure 1) were compiled for the glochidia of all available species. When possible ($n = 23$ species), glochidia were sampled from live, gravid females collected in Ontario by DFO staff and researchers in the Ackerman Laboratory at the University of Guelph following the methods of McNichols et al. 2011. Specifically, the shells of female mussels were opened gently and gills were flushed with water. Mature glochidia were collected, preserved in ethanol and measured to the nearest $0.01\ \mu\text{m}$ under a dissecting scope using Northern Eclipse image analysis software (Empix Inc. Mississauga, ON, Canada). When gravid females could not be found in the study area, glochidial measurements were determined from the published literature or were supplied by colleagues ($n = 13$ species) (Hoggarth 1999; M.C. Barnhart, Missouri State University, pers. comm.; A. Ford, U.S. Fish and Wildlife Service, pers. comm.; M. Hove, Missouri State University, pers. comm.; B. Sietman, Minnesota Department of Natural resources, pers. comm.). Information regarding the presence or absence of hooks was obtained from Clarke (1981), Hoggarth (1999), and Watters et al. (2009).

Measurements from all thirty-six unionid species were available for the analyses and characterized using discriminant function analysis (DFA) of glochidia height, hinge length, and

length (Table 1). DFA is a multivariate statistical method in which the user inputs raw data corresponding to known categories to build the model, and discriminant functions are created to maximize the among-groups differences (Quinn and Keough 2002).

Glochidial measurements from known species were used to build the DFA models. Raw measurements were log-transformed to meet the assumptions of normality, homogeneity of within-group (species) variance-covariance matrices, and absence of multivariate outliers (Quinn and Keough 2002). The model was then tested using a jackknife procedure, in which the observation being classified is omitted from the model calculation, thus removing an inherent bias in the classification procedure (Quinn and Keough 2002). The number of jackknife iterations was equal to the number of species included in the model. Five primary models were created – one which included all species that occur, or historically occurred, in (1) southern Ontario and one for each of the (2) Ausable, (3) Grand, (4) Sydenham, and (5) Thames rivers.

In addition to the five models described above, it was desirable to determine whether the separation of species with hooked and unhooked glochidia into separate models would increase classification success. The presence or absence of this coarse morphometric feature can often be readily distinguished and this has the potential to improve identification success. In order to assess the potential improvement to the model, two versions of the Southern Ontario model were created. The first model includes only species with hooked glochidia and the second model includes only those with unhooked glochidia (for a total of seven models). Classification success of individual species was then compared between the original "Southern Ontario" model and the separate hooked/unhooked versions.

3.0 RESULTS

3.1 GENERAL TRENDS

Two distinct groupings of species were observed in the graphs of the first and second discriminant functions for each model (Figures 2-6). Upon further inspection, these groupings represented "normally-sized" glochidia and micro-glochidia, respectively. When a species was misclassified, it was usually as another species within its grouping (Tables 2, 6, 8, 10, and 12).

The glochidia of some species were consistently misclassified in general. For the overall model, these included: *E. dilatata* (42.2% classification success), *E. triquetra* (40.0%), *L. fasciola* (45.5%), *L. siliquoidea* (16.8%), *L. recta* (40.0%), and *T. truncata* (37.5%). Upon examination of the graphs, the first and second discriminant functions of these species exhibited a high degree of overlap with those of other species (Figures 2-6).

The assumption of equal population covariance matrices was tested for all five DFA models using Box's M Test and was rejected in all instances. However, this test is very sensitive to large sample sizes (Quinn and Keough, 2009). Moreover, when "separate groups" covariance was used, the models changed very little, and this is thought to indicate that the assumptions have, more or less, been met. See relevant section for specific Box's M Test results.

3.2 SOUTHERN ONTARIO MODEL

The model's classification success was 74.3% (Table 2; Figure 2). Significant differences among group (species) means were detected for glochidia height, hinge length and length (Wilks' lambda = $W_{35,2992} = 0.033$, $p < 0.001$; $W_{35,2992} = 0.028$, $p < 0.001$; $W_{35,2992} = 0.021$, $p < 0.001$, respectively), which indicates that the model is effective at distinguishing between groups (species) based on these values. The null hypothesis of equal population covariance matrices was rejected (Box's M Test, $M = 5044.37$, $F_{204,20713} = 22.882$, $p < 0.001$).

The coefficients (x_1 , x_2 , x_3) and constant (b) associated with each species for the southern Ontario model used in the classification score equation can be found in Table 5.

When glochidia were separated into two models based on the presence or absence of hooks, the overall classification success of the unhooked glochidia model was reduced to 69.8%, whereas the model that included only hooked glochidia increased to 90.5%. However, changes in the classification success of individual species were minimal (Table 3; Table 4).

As indicated above, the "unhooked" model's classification success was 69.8% (Table 3; Figure 2). Significant differences among group means were detected for glochidia height, hinge length and length ($W_{24,2284} = 0.048$, $p < 0.001$; $W_{24,2284} = 0.056$, $p < 0.001$; $W_{24,2284} = 0.032$, $p < 0.001$, respectively), which indicates that the model is effective at distinguishing between groups based on these values. The null hypothesis of equal population covariance matrices was rejected ($M = 3112.34$, $F_{138,48282} = 21.662$, $p < 0.001$).

The classification success of the "hooked" glochidial model was 90.5% (Table 4; Figure 2). Significant differences among group means were detected for glochidia height, hinge length and length ($W_{10,708} = 0.092$, $p < 0.001$; $W_{10,708} = 0.032$, $p < 0.001$; $W_{10,708} = 0.211$, $p < 0.001$, respectively), which indicates that the model is effective at distinguishing between groups (species) based on these values. The null hypothesis of equal population covariance matrices was rejected ($M = 493.22$, $F_{60,3117} = 6.95$, $p < 0.001$).

3.3 AUSABLE RIVER MODEL

The model's classification success was 71.1% (Table 6; Figure 3). Significant differences among group means were detected for glochidia height, hinge length and length ($W = 0.040$, $p < 0.001$; $W = 0.038$, $p < 0.001$; $W = 0.036$, $p < 0.001$, respectively; $df = 24, 1844$), which indicates that the model is effective at distinguishing between groups (species) based on these values. The null hypothesis of equal population covariance matrices was rejected ($M = 2289$, $F_{144, 13046} = 14.5$, $p < 0.001$).

The coefficients and constant associated with each species for the Ausable River model can be found in Table 7.

3.4 GRAND RIVER MODEL

The model's classification success was 77.9% (Table 8; Figure 4). Significant differences among group means were detected for glochidia height, hinge length and length ($W_{30, 2752} = 0.030$, $p < 0.001$; $W_{30, 2752} = 0.026$, $p < 0.001$; $W_{30, 2752} = 0.020$, $p < 0.001$, respectively), which indicates that the model is effective at distinguishing between groups (species) based on these values. The null hypothesis of equal population covariance matrices was rejected ($M = 4777.27$, $F_{174, 17963} = 25.448$, $p < 0.001$).

The coefficients and constant associated with each species for the Grand River model can be found in Table 9.

3.5 SYDENHAM RIVER MODEL

The model's classification success was 75.1% (Table 10; Figure 5). Significant differences among group means were detected for glochidia height, hinge length and length ($W_{33, 2944} = 0.032$, $p < 0.001$; $W_{33, 2944} = 0.028$, $p < 0.001$; $W_{33, 2944} = 0.021$, $p < 0.001$, respectively), which indicates that the model is effective at distinguishing between groups (species) based on these values. The null hypothesis of equal population covariance matrices was rejected ($M = 4965.29$, $F_{198, 19633} = 23.169$, $p < 0.001$).

The coefficients and constant associated with each species for the Sydenham River model can be found in Table 11.

3.6 THAMES RIVER MODEL

The model's classification success was 74.8% (Table 12; Figure 6). Significant differences among group means were detected for glochidia height, hinge length and length ($W_{34, 2946} = 0.033$, $p < 0.001$; $W_{34, 2946} = 0.028$, $p < 0.001$; $W_{34, 2946} = 0.021$, $p < 0.001$, respectively), which

indicates that the model is effective at distinguishing between groups (species) based on these values. The null hypothesis of equal population covariance matrices was rejected ($M = 5011.43$, $F_{198,19633} = 23.384$, $p < 0.001$).

The coefficients and constant associated with each species for the Sydenham River model can be found in Table 13.

3.7 FURTHER ANALYSIS OF THE MODEL

To further investigate the effectiveness of the models at classifying unknown species of glochidia, the sensitivity of the algorithm for each species was determined for the Southern Ontario model. The sensitivity can be thought of as the probability that a glochidium of a given species will be classified as that species, and is equal to the classification success of a species. It can be calculated as:

$$\text{sensitivity} = \text{number of true positives} / [\text{true positives} + \text{false negatives}];$$

where a true positive is an incidence of a glochidium belonging to species "x" being classified as species "x"; a false negative can be thought of as the opposite: an incidence of a glochidium belonging to species "x" being classified as something other than species "x" (Sackett et al. 2000). In this case, the sensitivity of the algorithms for all the species in the model was quite variable (between 0.147 and 1.00), indicating that individuals belonging to a given species were not always identified correctly. For example, *L. siliquoidea* had a low sensitivity in the model (0.168), which suggests that glochidia belonging to this species have a high likelihood of not being identified correctly. See Table 2, 3, 4, 6, 8, 10, and 12 for classification success rates.

4.0 DISCUSSION

The southern Ontario and river-specific DFA models were effective in classifying glochidia in this study and could be used in the future to identify glochidia collected from the water column or glochidia that have become encysted on wild-caught fish. Knowledge of the unionid species that infested a particular fish species may provide insight into mechanisms promoting infestation (e.g., encounter of glochidia in the water column, attempted predation of conglutinates; Barnhart et al. 2008). In addition, observation of natural infestations could be used as supporting evidence for host fish relationships determined in the laboratory, or as starting points for selecting pairings to examine in the laboratory. We recommend caution when using these models to imply direct host relationships as the identification of glochidial attachment does not imply successful transformation (Jansen et al. 2001; see discussion in Haag and Warren 2003).

Our results indicate that the identification of glochidia using shell morphometrics is a reasonable and worthwhile endeavour with overall success rates between 71.1% and 77.9 %. This success rate is far higher than would be expected by chance alone.

However, it is clear that not all species can be readily identified using this technique as the glochidia of some species were consistently misclassified. For instance, the classification success, or sensitivity, of *L. siliquoidea* was the lowest in every model (16-25 %), indicating that these models are not effective at identifying these glochidia in the water column, nor useful at discriminating among species of *Lampsilis* species in general (e.g., *L. cardium*, *L. fasciola*, *L. siliquoidea*) or between species in the genus *Truncilla* (e.g., *T. truncata* and *T. donaciformis*). These misidentifications resulted from a high degree of overlap between the morphological characteristics of these species. Our attempt to improve classification success by pre-sorting glochidia on the basis of the presence/absence of hooks was not successful indicating that there is little morphological overlap among these two groups in the three axes examined. Although the overall success of the hooked model increased, the individual success rates changed very little indicating these species were well sorted in both models and that the change in success reflected the elimination of the unhooked group where most morphological overlap exists. In general we do not recommend the use of separate models on the basis of hook presence/absence because of the lack of increased discrimination success and the potential of introducing additional error into the classification process during the pre-sorting phase (e.g., broken or missing hooks). The only species to show a large change in classification success in the separate models was the unhooked *Epioblasma torulosa rangiana* which saw a 10% increase in classification success when isolated from the hooked specimens. This represents the only case where an unhooked species is often misidentified as a hooked species or vice versa. Studies designed to target glochidia of this endangered species may wish to employ a model pre-sorting for the presence/absence of hooks.

As would be expected, generally, the greater the number of species that are included in the model, the lower the classification success of unknown glochidia. The exception to this is the Ausable River model, which included the fewest species ($n = 25$) and had the lowest classification success (71.1%). This is likely a product of the constellation of species included in the model; for instance, it is difficult to distinguish between species within the *Lampsilis* and *Epioblasma* genera and both are heavily represented in the Ausable River model. In spite of this, we recommend that the river-specific model be used if glochidial origin is known. In addition, it would be beneficial to narrow the potential list of species that a glochidium could belong to, if possible, in order to improve accuracy of the results.

4.1 APPLICATION OF THE MODEL

The purpose of the present study was to develop a statistical model that could be used to identify unknown glochidia from the waters of southern Ontario. We have demonstrated that it is possible to assess the identification of most glochidia from this region using a few simple

morphological measures. Using the classification functions derived through this analysis it is possible to develop a tool for the identification of unknown glochidia collected anywhere in southern Ontario. The identity of a given glochidium can be determined by inserting its log-transformed dimensions (i.e., hinge length, length, and height in μm) into the classification functions for all of the species within the appropriate model. Within this function, each log-transformed independent variable (i.e., hinge length, length, and height) is multiplied by its corresponding coefficient (x_1 , x_2 , or x_3) and these values are then summed along with the constant (b ; Table 5) to obtain a classification score (IBM Support, 2011) such that:

$$\text{Classification score} = (\log \text{ height of observation} * x_1) + (\log \text{ hinge length of observation} * x_2) + (\log \text{ length of observation} * x_3) + b$$

The unknown glochidium should then be assigned to the species for which the highest classification score is obtained.

The associated probability that the unknown glochidium belongs to a given group can then be calculated as:

$$P(\text{group} = i) = \frac{e^{C_i - C_{\max}}}{\sum_{i=1}^k e^{C_i - C_{\max}}}$$

where C_i is the classification score for group i , C_{\max} is the maximum classification score for the glochidium, and $\sum_{i=1}^k e^{C_i - C_{\max}}$ is the total sum of $e^{C_i - C_{\max}}$ results over all (k) groups (IBM Support, 2011).

5.0 CONCLUSIONS

The use of DFA models to determine the identity of unknown glochidia represents a powerful, cost effective tool for use in the management of species at risk. The classification models presented in this report can be used across southern Ontario to identify glochidia collected from a variety of sources (drift samples, fish collections, etc.). Where available we recommend the use of watershed specific models however the more general southern Ontario model can be widely applied across the region.

6.0 ACKNOWLEDGEMENTS

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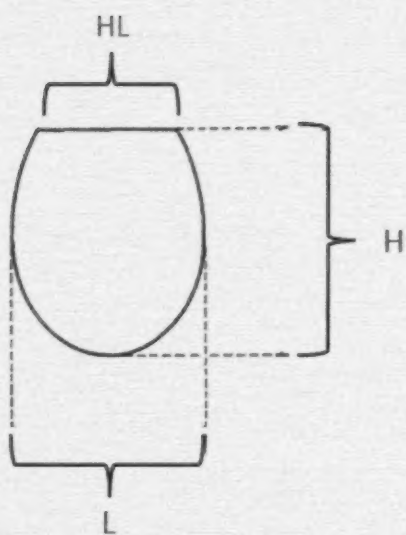


Figure 1. A description of the variables that were measured: Hinge Length (HL), Length (L) and Height (H) (modified from Hoggarth 1999)

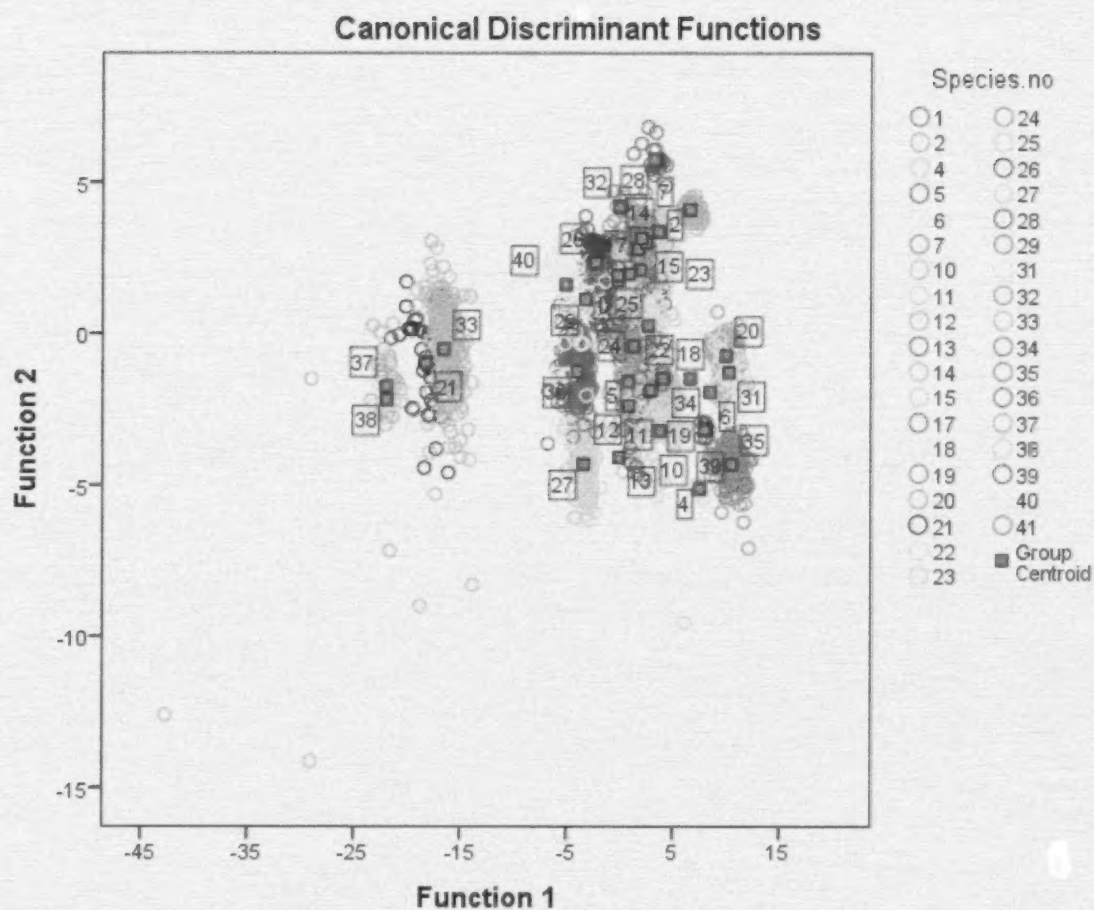


Figure 2. Plot of canonical discriminant functions 1 and 2 for the species included in the southern Ontario regional model. Numbers correspond to species numbers in tables.

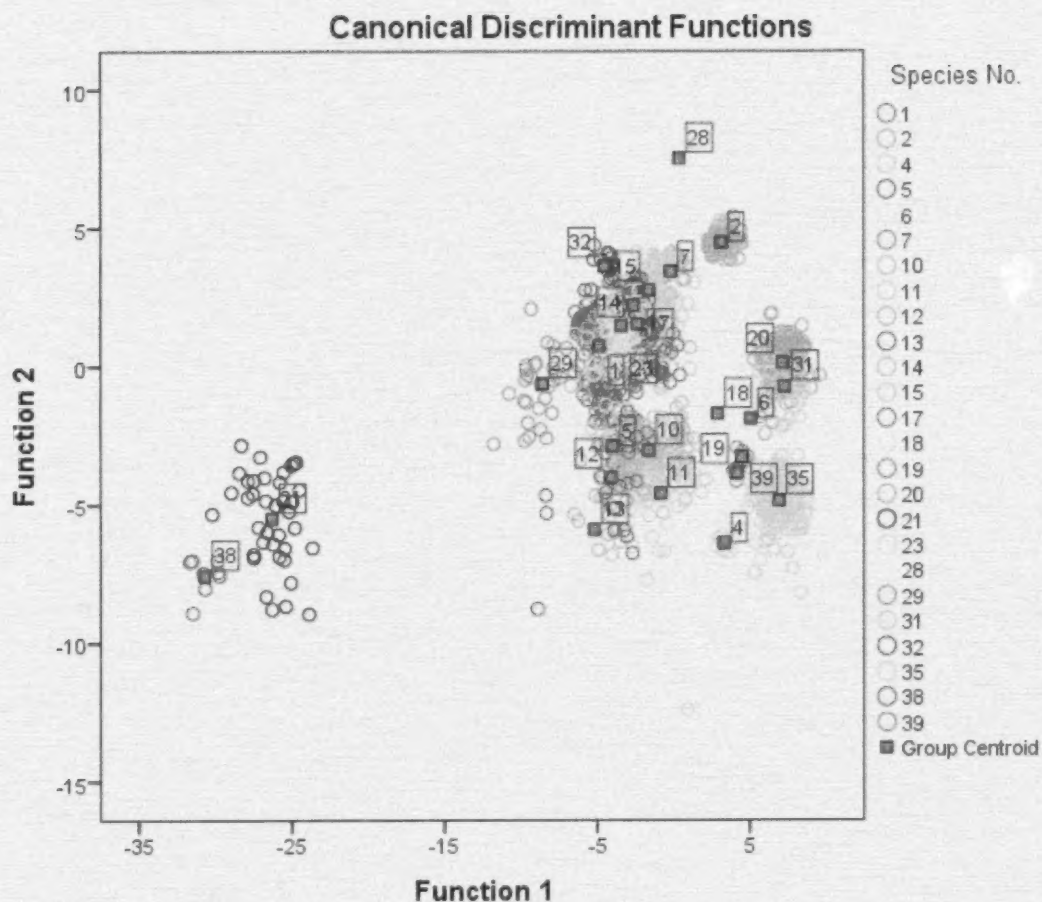


Figure 3. Plot of canonical discriminant functions 1 and 2 for the species included in the Ausable River model. Numbers correspond to species numbers in tables.

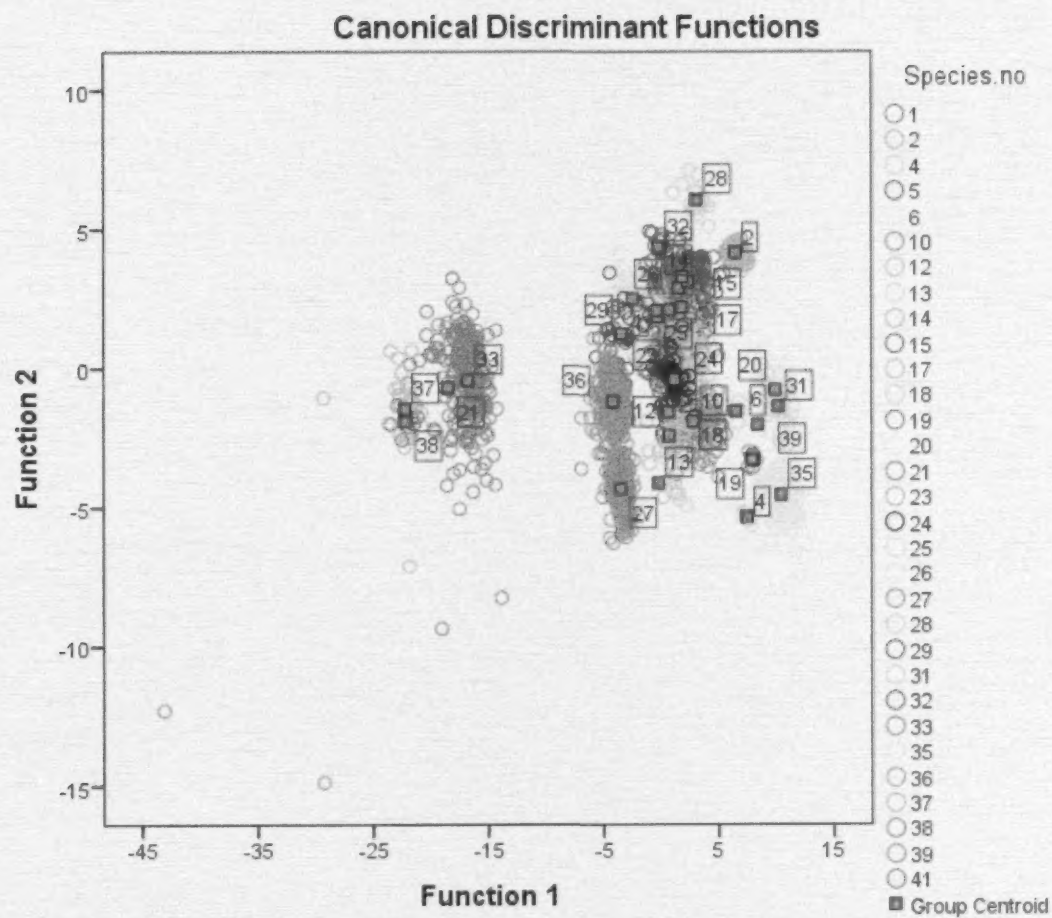


Figure 4. Plot of canonical discriminant functions 1 and 2 for the species included in the Grand River model. Numbers correspond to species numbers in tables.

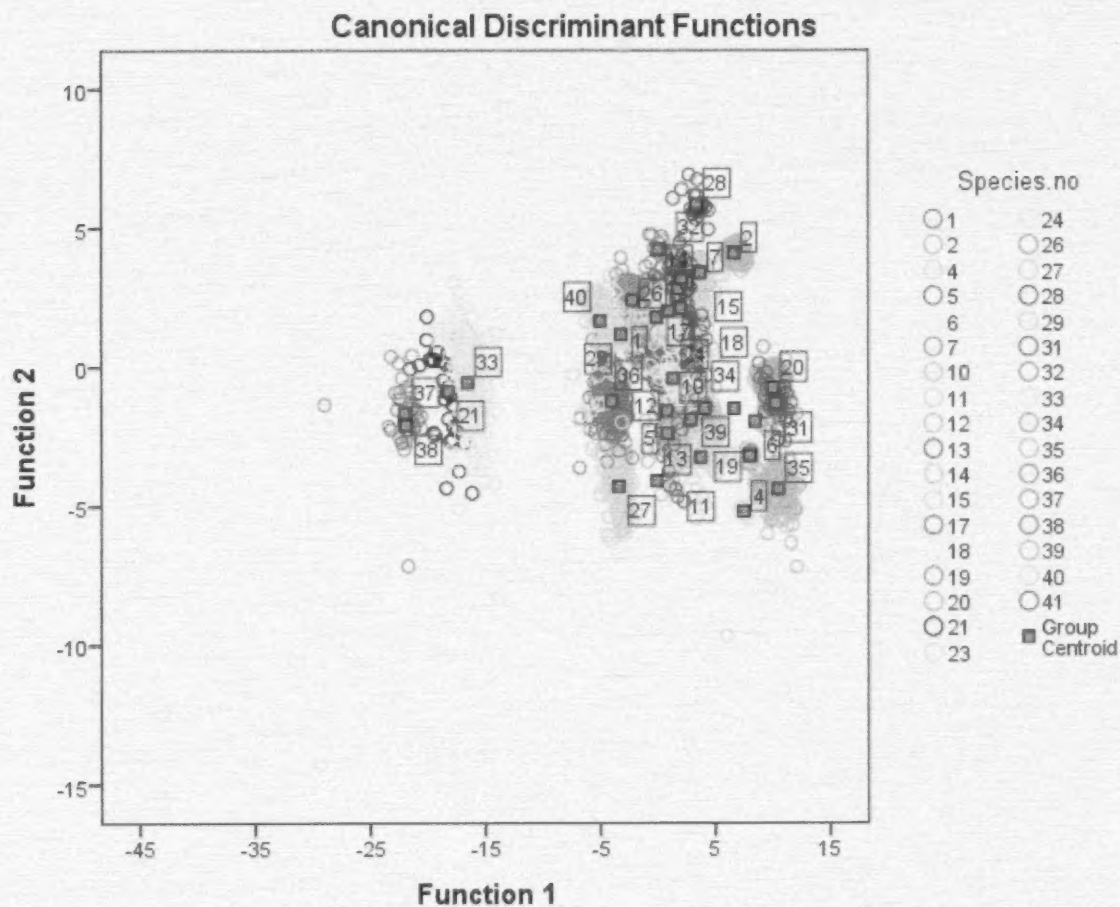


Figure 5. Plot of canonical discriminant functions 1 and 2 for the species included in the Sydenham River model. Numbers correspond to species numbers in tables.

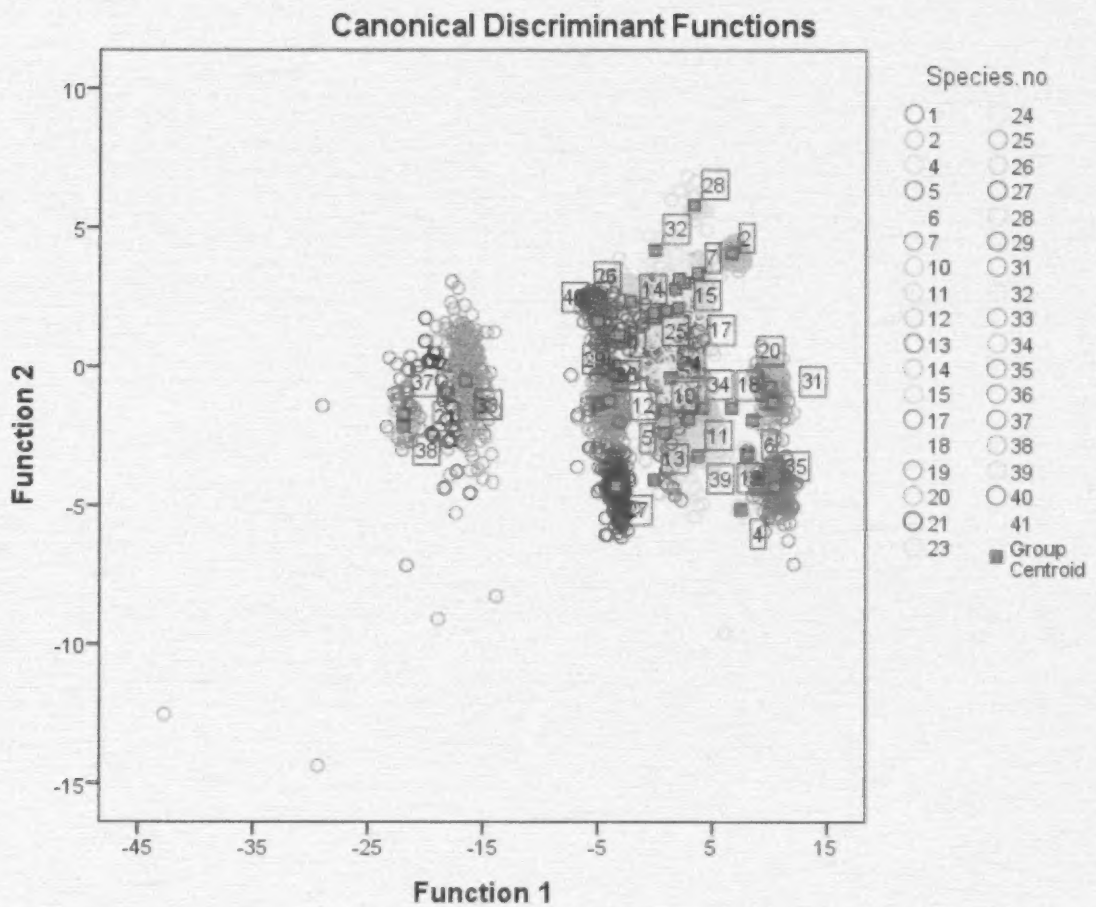


Figure 6. Plot of canonical discriminant functions 1 and 2 for the species included in the Thames River model. Numbers correspond to species numbers in tables.

Table 1. Summary of species included in Discriminant Function Analysis (DFA) model. AR = Ausable River; GR = Grand River; SR = Sydenham River; TR = Thames River

Species	n _{total}	mean \pm S.D. (μ m)				DFA model				
		shell height	hinge length	shell length	Hooks	AR	GR	SR	TR	SW Ontario
1 Mucket, <i>Actinonaias ligamentina</i>	166	244.65 \pm 11.62	105.24 \pm 15.67	210.17 \pm 9.12	x	✓	✓	✓	✓	✓
2 Elktoe, <i>Alasmidonta marginata</i>	109	379.66 \pm 10.25	140.23 \pm 4.75	324.82 \pm 10.10	✓	✓	✓	✓	✓	✓
4 Slippershell, <i>Alasmidonta viridis</i> ¹	7	250.29 \pm 4.35	251.14 \pm 5.84	306.86 \pm 7.95	✓	✓	✓	✓	✓	✓
5 Threeridge, <i>Amblema plicata</i> ²	20	221.22 \pm 9.69	136.96 \pm 11.74	209.29 \pm 8.15	x	✓	✓	✓	✓	✓
6 Cylindrical Papershell, <i>Anodontoides ferussacianus</i> ¹	7	323.86 \pm 9.47	234.29 \pm 2.93	323.00 \pm 2.52	✓	✓	✓	✓	✓	✓
7 Purple Wartyback, <i>Cyclonaias tuberculata</i> ³	13	325.05 \pm 9.47	123.88 \pm 5.23	264.35 \pm 5.38	x	✓	x	✓	✓	✓
10 Spike, <i>Elliptio dilatata</i>	128	239.13 \pm 14.82	158.06 \pm 16.76	240.04 \pm 21.41	x	✓	✓	✓	✓	✓
11 Northern Riffleshell, <i>Epioblasma torulosa rangiana</i>	47	231.53 \pm 18.72	179.32 \pm 24.40	249.81 \pm 21.91	x	✓	x	✓	✓	✓

Species	n _{total}	mean ± S.D. (µm)			DFA model					
		shell height	hinge length	shell length	Hooks	AR	GR	SR	TR	SW Ontario
12 Snuffbox, <i>Epioblasma triquetra</i> ⁶	40	209.88 ± 14.16	141.76 ± 14.51	211.23 ± 11.14	x	✓	✓	✓	✓	✓
13 Wabash Pigtoe, <i>Fusconaia flava</i>	9	189.69 ± 21.36	151.49 ± 23.74	192.38 ± 23.44	x	✓	✓	✓	✓	✓
14 Plain Pocketbook, <i>Lampsilis cardium</i>	225	280.33 ± 9.80	110.46 ± 11.16	239.66 ± 11.97	x	✓	✓	✓	✓	✓
15 Wavyrayed lampmussel, <i>Lampsilis fasciola</i> ⁶	222	300.40 ± 23.86	116.62 ± 9.16	247.88 ± 18.57	x	✓	✓	✓	✓	✓
17 Fatmucket, <i>Lampsilis siliquoidea</i>	143	277.72 ± 18.70	118.40 ± 15.16	237.95 ± 8.14	x	✓	✓	✓	✓	✓
18 White Heelsplitter, <i>Lasmigona complanata</i> ¹	6	300.17 ± 7.08	201.00 ± 5.10	292.83 ± 2.79	✓	✓	✓	✓	✓	✓
19 Creek Heelsplitter, <i>Lasmigona compressa</i> ¹	5	285.80 ± 1.92	233.60 ± 3.36	322.60 ± 4.39	✓	✓	✓	✓	✓	✓
20 Flutedshell, <i>Lasmigona costata</i>	315	383.36 ± 13.83	250.06 ± 10.91	348.99 ± 12.30	✓	✓	✓	✓	✓	✓
21 Fragile Papershell, <i>Leptodea fragilis</i> ²	38	86.07 ± 6.38	37.06 ± 5.97	70.66 ± 6.76	x	✓	✓	✓	✓	✓
22 Eastern Pondmussel, <i>Ligumia nasuta</i> ⁶	47	273.14 ± 30.08	142.06 ± 10.30	235.95 ± 12.70	x	x	✓	x	x	✓

Species	n _{total}	mean ± S.D. (µm)			DFA model					
		shell height	hinge length	shell length	Hooks	AR	GR	SR	TR	SW Ontario
23 Black Sandshell, <i>Ligumia recta</i>	50	268.59 ± 16.45	113.73 ± 8.62	219.08 ± 13.58	x	✓	✓	✓	✓	✓
24 Threehorn Wartyback, <i>Obliquaria reflexa</i> ⁶	31	233.42 ± 10.79	128.40 ± 9.52	224.89 ± 10.30	x	x	✓	✓	✓	✓
25 Hickorynut, <i>Obovaria olivaria</i> ^{1, 6}	3	257.67 ± 3.51	107.67 ± 2.08	202.00 ± 4.00	x	x	✓	x	✓	✓
26 Round Hickorynut, <i>Obovaria subrotunda</i> ⁶	97	228.87 ± 9.87	88.66 ± 7.07	185.30 ± 7.79	x	x	✓	✓	✓	✓
27 Round Pigtoe, <i>Pleurobema sintoxia</i> ^{5, 6}	191	154.92 ± 7.13	119.81 ± 7.67	159.76 ± 5.99	x	x	✓	✓	✓	✓
28 Pink Heelsplitter, <i>Potamilus alatus</i>	20	405.65 ± 9.98	118.89 ± 7.14	239.81 ± 12.71	✓	✓	✓	✓	✓	✓
29 Kidneyshell, <i>Ptychobranhus fasciolaris</i> ⁶	33	201.39 ± 17.12	87.30 ± 7.60	176.10 ± 14.38	x	✓	✓	✓	✓	✓
31 Giant Floater, <i>Pyganadon grandis</i>	30	369.46 ± 14.62	257.51 ± 22.60	359.15 ± 14.48	✓	✓	✓	✓	✓	✓
32 Pimpleback, <i>Quadrula pustulosa</i> ⁴	14	276.36 ± 9.56	90.93 ± 3.93	218.43 ± 8.79	x	✓	✓	✓	✓	✓
33 Mapleleaf, <i>Quadrula quadrula</i> ^{5, 6}	259	91.26 ± 9.59	38.19 ± 5.94	82.88 ± 6.59	x	x	✓	✓	✓	✓

Species	n _{total}	mean ± S.D. (µm)			DFA model					
		shell height	hinge length	shell length	Hooks	AR	GR	SR	TR	SW Ontario
34 Salamander Mussel, <i>Simpsonaias ambigua</i> ^{1, 6}	6	260.83 ± 3.97	167.83 ± 2.22	254.83 ± 2.56	✓	x	x	✓	✓	✓
35 Creeper, <i>Strophitus undulatus</i>	207	305.43 ± 13.32	294.91 ± 17.10	366.77 ± 20.70	✓	✓	✓	✓	✓	✓
36 Lilliput, <i>Toxolasma parvum</i> ⁶	318	173.48 ± 8.15	95.88 ± 6.32	161.48 ± 6.38	x	x	✓	✓	✓	✓
37 Fawnsfoot, <i>Truncilla donaciformis</i> ⁶	31	68.23 ± 6.18	29.91 ± 3.76	56.58 ± 4.71	x	x	✓	✓	✓	✓
38 Deertoe, <i>Truncilla truncata</i>	8	67.29 ± 3.3	30.85 ± 1.48	56.04 ± 3.99	x	✓	✓	✓	✓	✓
39 Paper Pondshell, <i>Utterbackia imbecillis</i> ¹	7	301.86 ± 5.79	247.43 ± 5.88	304.86 ± 5.70	✓	✓	✓	✓	✓	✓
40 Rayed Bean, <i>Villosa fabalis</i> ⁶	132	191.08 ± 16.27	76.57 ± 9.13	156.89 ± 10.52	x	x	x	✓	✓	✓
41 Rainbow, <i>Villosa iris</i> ⁶	44	303.80 ± 11.43	115.62 ± 9.12	234.76 ± 11.24	x	x	✓	✓	✓	✓
TOTAL						25	32	34	35	36

✓ = species included in model, x = species not included in model

n_{total} refers to the total number of individual glochidia used to create each model

¹ Hoggarth, 1999

² A. Ford, U.S. Fish and Wildlife Service, pers. comm., June 2012

³ M.C. Barnhart, Missouri State University, pers. comm., June 2012

⁴ M. Hove, Missouri State University, pers. comm., June 2012

⁵ B.E. Sietman, Minnesota Department of Natural Resources, pers. comm., May 2014

⁶ Species assessed as Endangered, Threatened, or Special Concern by COSEWIC.

Table 2. Discriminant Function Analysis (DFA) model with all Southern Ontario species included

	Species	N	% correct	Misclassified as (%)
2	Elktoe, <i>A. marginata</i>	109	100	--
28	Pink Heelsplitter, <i>P. alatus</i>	20	100	--
32	Pimpleback, <i>Q. pustulosa</i>	14	100	--
7	Purple Wartyback, <i>C. tuberculata</i>	13	100	--
6	Cylindrical Papershell, <i>A. ferussacianus</i>	7	100	--
4	Slippershell, <i>A. viridis</i>	7	100	--
39	Paper Pondshell, <i>U. imbecillis</i>	7	100	--
18	White Heelsplitter, <i>L. complanata</i>	6	100	--
34	Salamander Mussel, <i>S. ambigua</i>	6	100	--
19	Creek Heelsplitter, <i>L. compressa</i>	5	100	--
25	Hickorynut, <i>Obovaria olivaria</i>	3	100	--
27	Round Pigtoe, <i>Pleurobema sintoxia</i>	191	96.9	<i>T. parvum</i> (3.1)
36	Lilliput, <i>T. parvum</i>	318	94.7	<i>Pleurobema sintoxia</i> (1.6), <i>P. fasciolaris</i> (2.2), <i>V. fabalis</i> (1.6)
35	Creeper, <i>S. undulatus</i>	207	92.8	<i>A. viridis</i> (0.5), <i>L. compressa</i> (1.4), <i>U. imbecillis</i> (5.3)
33	Mapleleaf, <i>Quadrula quadrula</i>	259	89.2	<i>L. fragilis</i> (9.7), <i>T. donaciformis</i> (0.8), <i>T. truncata</i> (0.4)
20	Flutedshell, <i>L. costata</i>	315	86.3	<i>A. ferussacianus</i> (1.0), <i>P. grandis</i> (12.7)

	Species	N	% correct	Misclassified as (%)
26	Round Hickorynut, <i>O. subrotunda</i>	97	84.5	<i>A. ligamentina</i> (6.2), <i>O. oliviara</i> (1.0), <i>P. fasciolaris</i> (7.2), <i>V. fabalis</i> (1.0)
40	Rayed Bean, <i>V. fabalis</i>	132	75.8	<i>O. oliviara</i> (0.8), <i>O. subrotunda</i> (1.5), <i>P. sintoxia</i> (0.8), <i>P. fasciolaris</i> (10.6), <i>T. parvum</i> (10.6)
14	Plain Pocketbook, <i>L. cardium</i>	225	74.7	<i>A. ligamentina</i> (1.8), <i>L. fasciola</i> (6.7), <i>L. siliquoidea</i> (4.9), <i>L. nasuta</i> (6.2), <i>L. recta</i> (3.1), <i>O. oliviara</i> (1.8), <i>S. ambigua</i> (0.4), <i>V. iris</i> (0.4)
21	Fragile Papershell, <i>L. fragilis</i>	38	73.7	<i>Q. quadrula</i> (21.1), <i>T. donaciformis</i> (5.3)
24	Threehorn Wartyback, <i>O. reflexa</i>	31	74.2	<i>A. ligamentina</i> (3.2), <i>A. plicata</i> (9.7), <i>E. dilatata</i> (6.5), <i>L. siliquoidea</i> (3.2), <i>L. nasuta</i> (3.2)
41	Rainbow, <i>V. iris</i>	44	72.7	<i>C. tuberculata</i> (2.3), <i>L. cardium</i> (2.3), <i>L. fasciola</i> (15.9), <i>L. nasuta</i> (4.5), <i>Q. pustulosa</i> (2.3)
31	Giant Floater, <i>P. grandis</i>	30	70.0	<i>A. ferussacianus</i> (6.7), <i>L. costata</i> (23.3)
11	Northern Riffleshell, <i>E. t. rangiana</i>	47	66.0	<i>A. viridis</i> (2.1), <i>A. plicata</i> (2.1), <i>E. dilatata</i> (12.8), <i>E. triquetra</i> (2.1), <i>F. flava</i> (2.1), <i>L. complanata</i> (2.1), <i>L. compressa</i> (4.3), <i>O. reflexa</i> (2.1), <i>S. ambigua</i> (4.3)
22	Eastern Pondmussel, <i>L. nasuta</i>	47	63.8	<i>A. plicata</i> (4.3), <i>C. tuberculata</i> (4.3), <i>E. dilatata</i> (2.1), <i>E. triquetra</i> (2.1), <i>L. recta</i> (2.1), <i>O. reflexa</i> (19.1), <i>V. iris</i> (2.1)
37	Fawnsfoot, <i>Truncilla donaciformis</i>	31	58.1	<i>L. fragilis</i> (3.2), <i>T. truncata</i> (38.7)
13	Wabash Pigtoe, <i>Fusconaia flava</i>	9	55.6	<i>E. t. rangiana</i> (11.1), <i>P. sintoxia</i> (33.3)
5	Threeridge, <i>A. plicata</i>	20	55.0	<i>E. dilatata</i> (5.0), <i>E. triquetra</i> (25.0), <i>O. reflexa</i> (15.0)
1	Mucket, <i>A. ligamentina</i>	166	53.0	<i>A. plicata</i> (7.8), <i>E. dilatata</i> (0.6), <i>L. siliquoidea</i> (0.6), <i>L. nasuta</i> (4.2), <i>L. recta</i> (4.8), <i>O. reflexa</i> (6.6), <i>O. oliviara</i> (9.0), <i>O. subrotunda</i> (5.4), <i>Q. pustulosa</i> (7.2), <i>V. iris</i> (0.6)

	Species	N	% correct	Misclassified as (%)
29	Kidneyshell, <i>P. fasciolaris</i>	33	48.5	<i>A. ligamentina</i> (12.1), <i>O. subrotunda</i> (9.1), <i>T. parvum</i> (9.1), <i>V. fabalis</i> (21.2)
15	Wavyrayed Lamprussel, <i>L. fasciola</i>	222	45.9	<i>A. ligamentina</i> (2.3), <i>A. marginata</i> (1.4), <i>A. plicata</i> (0.5), <i>C. tuberculata</i> (22.1), <i>L. cardium</i> (5.4), <i>L. siliquoidea</i> (3.2), <i>L. nasuta</i> (1.4), <i>L. recta</i> (9.9), <i>O. reflexa</i> (0.9), <i>O. olivaria</i> (2.3), <i>O. subrotunda</i> (0.5), <i>P. alatus</i> (0.5), <i>S. ambigua</i> (0.5), <i>V. iris</i> (3.6)
10	Spike, <i>E. dilatata</i>	128	42.2	<i>A. plicata</i> (16.4), <i>E. t. rangiana</i> (11.7), <i>E. triquetra</i> (3.9), <i>L. cardium</i> (0.8), <i>L. siliquoidea</i> (0.8), <i>L. complanata</i> (0.8), <i>L. nasuta</i> (0.8), <i>O. reflexa</i> (1.6), <i>O. olivaria</i> (1.6), <i>S. ambigua</i> (19.5)
23	Black Sandshell, <i>L. recta</i>	50	40.0	<i>A. ligamentina</i> (10.0), <i>A. plicata</i> (2.0), <i>L. cardium</i> (6.0), <i>L. fasciola</i> (2.0), <i>L. siliquoidea</i> (10.0), <i>L. nasuta</i> (4.0), <i>O. olivaria</i> (18.0), <i>Q. pustulosa</i> (2.0), <i>V. iris</i> (6.0)
12	Snuffbox, <i>E. triquetra</i>	40	40.0	<i>A. ligamentina</i> (2.5), <i>A. plicata</i> (27.5), <i>E. dilatata</i> (7.5), <i>F. flava</i> (12.5), <i>O. reflexa</i> (10.0)
38	Deertoe, <i>T. truncata</i>	8	37.5	<i>T. donaciformis</i> (62.5)
17	Fatmucket, <i>L. siliquoidea</i>	143	16.8	<i>A. ligamentina</i> (0.7), <i>C. tuberculata</i> (0.7), <i>E. dilatata</i> (2.8), <i>E. triquetra</i> (0.7), <i>L. cardium</i> (53.1), <i>L. fasciola</i> (2.1), <i>L. nasuta</i> (14.7), <i>L. recta</i> (0.7), <i>O. reflexa</i> (4.9), <i>V. iris</i> (2.8)
	TOTAL	3028	74.3	

Table 3. Discriminant Function Analysis (DFA) model including only Southern Ontario species with unhooked glochidia

Species	N	% correct classification in unhooked model	% correct classification in all-species model	% change
7 Purple Wartyback, <i>C. tuberculata</i>	13	100	100	0
25 Hickorynut, <i>Obovaria olivaria</i>	3	100	100	0
32 Pimpleback, <i>Q. pustulosa</i>	14	100	100	0
27 Round Pigtoe, <i>P. sintoxia</i>	191	96.9	96.9	0
36 Lilliput, <i>T. parvum</i>	318	94.7	94.7	0
33 Mapleleaf, <i>Q. quadrula</i>	259	89.2	89.2	0
26 Round Hickorynut, <i>O. subrotunda</i>	97	84.5	84.5	0
11 Northern Riffleshell, <i>E. t. rangiana</i>	47	76.6	66	10.6
40 Rayed Bean, <i>V. fabalis</i>	132	76.5	75.8	0.7
14 Plain Pocketbook, <i>L. cardium</i>	225	74.7	74.7	0
24 Threehorn Wartyback, <i>O. reflexa</i>	31	74.2	74.2	0
21 Fragile Papershell, <i>L. fragilis</i>	38	73.7	73.7	0
41 Rainbow, <i>V. iris</i>	44	72.7	72.7	0
22 Eastern Pondmussel, <i>L. nasuta</i>	47	63.8	63.8	0
37 Fawnsfoot, <i>Truncilla donaciformis</i>	31	58.1	58.1	0

	Species	N	% correct classification in unhooked model	% correct classification in all-species model	% change
13	Wabash Pigtoe, <i>Fusconaia flava</i>	9	55.6	55.6	0
1	Mucket, <i>A. ligamentina</i>	166	53.6	53	0.6
5	Threeridge, <i>A. plicata</i>	20	50.0	55	-5
10	Spike, <i>E. dilatata</i>	128	49.2	42.2	7
29	Kidneyshell, <i>P. fasciolaris</i>	33	48.5	48.5	0
15	Wavyrayed Lampmussel, <i>L. fasciola</i>	222	45.0	45.9	-0.9
12	Snuffbox, <i>E. triquetra</i>	40	40.0	40	0
23	Black Sandshell, <i>L. recta</i>	50	40.0	40	0
38	Deertoe, <i>T. truncata</i>	8	37.5	37.5	0
17	Fatmucket, <i>L. siliquoidea</i>	143	16.8	16.8	0.7

Table 4. Discriminant Function Analysis (DFA) model including only Southern Ontario species with hooked glochidia

Species	N	% correct classification in hooked model	% correct classification in all-species model	% change
2 Elktoe, <i>A. marginata</i>	109	100	100	0
4 Slippershell, <i>A. viridis</i>	7	100	100	0
6 Cylindrical Papershell, <i>A. ferussacianus</i>	7	100	100	0
18 White Heelsplitter, <i>L. complanata</i>	6	100	100	0
19 Creek Heelsplitter, <i>L. compressa</i>	5	100	100	0
28 Pink Heelsplitter, <i>P. alatus</i>	20	100	100	0
34 Salamander Mussel, <i>S. ambigua</i>	6	100	100	0
39 Paper Pondshell, <i>U. imbecillis</i>	7	100	100	0
35 Creeper, <i>S. undulatus</i>	207	92.8	92.8	0
20 Flutedshell, <i>L. costata</i>	315	85.1	86.3	-1.2
31 Giant Floater, <i>P. grandis</i>	30	80.0	70	10

Table 5. Classification Function Coefficients for the "Southern Ontario" regional model

		Classification Function Coefficients			
		x ₁	x ₂	x ₃	b
Species		Log Height	Log Hinge Length	Log Length	(Constant)
1	Mucket, <i>A. ligamentina</i>	978.821	470.843	2083.313	-4066.232
2	Elktoe, <i>A. marginata</i>	1057.628	485.215	2266.310	-4734.156
4	Slippershell, <i>A. viridis</i>	856.151	679.936	2257.325	-4652.907
5	Threeridge, <i>A. plicata</i>	923.087	554.904	2071.438	-4081.332
6	Cylindrical Papershell, <i>A. ferussacianus</i>	966.340	643.609	2236.740	-4785.296
7	Purple Wartyback, <i>C. tuberculata</i>	1044.672	481.322	2163.494	-4439.300
10	Spike, <i>E. dilatata</i>	918.029	576.112	2136.803	-4268.890
11	Northern Riffleshell, <i>E. t. rangiana</i>	886.868	608.462	2161.086	-4324.911
12	Snuffbox, <i>E. triquetra</i>	894.080	565.296	2088.575	-4075.724
13	Wabash Pigtoe, <i>Fusconaia flava</i>	869.356	599.995	2031.316	-3962.089
14	Plain Pocketbook, <i>L. cardium</i>	1005.773	464.707	2141.728	-4256.253
15	Wavyrayed Lampmussel, <i>L. fasciola</i>	1026.473	474.592	2141.486	-4326.932
17	Fatmucket, <i>L. siliquoidea</i>	999.891	485.572	2130.935	-4259.102
18	White Heelsplitter, <i>L. complanata</i>	962.406	613.905	2200.934	-4617.003

		Classification Function Coefficients			
		x ₁	x ₂	x ₃	b
	Species	Log Height	Log Hinge Length	Log Length	(Constant)
19	Creek Heelsplitter, <i>L. compressa</i>	907.699	647.216	2270.268	-4732.341
20	Flutedshell, <i>L. costata</i>	1022.590	646.589	2242.648	-4950.578
21	Fragile Papershell, <i>L. fragilis</i>	818.945	343.514	1648.664	-2586.652
22	Eastern Pondmussel, <i>L. nasuta</i>	986.336	542.975	2104.095	-4283.492
23	Black Sandshell, <i>L. recta</i>	1008.567	487.409	2078.590	-4160.590
24	Threehorn Wartyback, <i>O. reflexa</i>	931.052	524.631	2121.662	-4153.202
25	Hickorynut, <i>Obovaria olivaria</i>	1013.281	483.984	2033.909	-4061.224
26	Round Hickorynut, <i>O. subrotunda</i>	987.417	441.320	2024.452	-3893.046
27	Round Pigtoe, <i>Pleurobema sintoxia</i>	832.913	564.128	1974.063	-3676.089
28	Pink Heelsplitter, <i>P. alatus</i>	1177.834	474.434	2030.961	-4447.660
29	Kidneyshell, <i>P. fasciolaris</i>	940.780	448.163	2020.400	-3788.498
31	Giant Floater, <i>P. grandis</i>	996.375	651.886	2271.676	-4969.784
32	Pimpleback, <i>Q. pustulosa</i>	1030.805	420.907	2098.698	-4128.339
33	Mapleleaf, <i>Quadrula quadrula</i>	797.866	329.575	1755.765	-2725.304
34	Salamander Mussel, <i>S. ambigua</i>	940.246	583.984	2154.014	-4380.724

		Classification Function Coefficients			
		x_1	x_2	x_3	b
	Species	Log Height	Log Hinge Length	Log Length	(Constant)
35	Creeper, <i>S. undulatus</i>	896.049	696.581	2321.119	-4951.926
36	Lilliput, <i>T. parvum</i>	891.166	492.766	1981.356	-3676.353
37	Fawnsfoot, <i>Truncilla donaciformis</i>	776.168	319.113	1566.093	-2320.520
38	Deertoe, <i>T. truncata</i>	772.000	331.748	1557.828	-2317.145
39	Paper Pondshell, <i>U. imbecillis</i>	946.970	669.920	2203.115	-4715.678
40	Rayed Bean, <i>V. fabalis</i>	951.633	425.738	1962.664	-3642.373
41	Rainbow, <i>V. iris</i>	1047.686	478.598	2097.453	-4282.927

Table 6. Discriminant Function Analysis (DFA) model including only Ausable River species

	Species	n	% correct	Misclassified as (%)
2	Elktoe, <i>A. marginata</i>	109	100	--
28	Pink Heelsplitter, <i>P. alatus</i>	20	100	--
32	Pimpleback, <i>Q. pustulosa</i>	14	100	--
7	Purple Wartback, <i>C. tuberculata</i>	13	100	--
38	Deertoe, <i>T. truncata</i>	8	100	--
6	Cylindrical Papershell, <i>A. ferussacianus</i>	7	100	--
4	Slippershell, <i>A. viridis</i>	7	100	--
39	Paper Pondshell, <i>U. imbecillis</i>	7	100	--
18	White Heelsplitter, <i>L. complanata</i>	6	100	--
19	Creek Heelsplitter, <i>L. compressa</i>	5	100	--
21	Fragile Papershell, <i>L. fragilis</i>	38	97.4	<i>T. truncata</i> (2.6)
35	Creeper, <i>S. undulatus</i>	207	91.8	<i>A. viridis</i> (1.4), <i>L. compressa</i> (1.4), <i>U. imbecillis</i> (5.3)
29	Kidneyshell, <i>P. fasciolaris</i>	33	87.9	<i>A. ligamentina</i> (12.1)
20	Flutedshell, <i>L. costata</i>	315	85.7	<i>A. ferussacianus</i> (1.6), <i>P. grandis</i> (12.7)

	Species	n	% correct	Misclassified as (%)
13	Wabash Pigtoe, <i>F. flava</i>	9	77.8	<i>E. t. rangiana</i> (11.1), <i>E. triquetra</i> (11.1)
14	Plain Pocketbook, <i>L. cardium</i>	225	74.7	<i>A. ligamentina</i> (2.2), <i>L. fasciola</i> (6.2), <i>L. siliquoidea</i> (9.8), <i>L. complanata</i> (0.9), <i>L. recta</i> (6.2)
1	Mucket, <i>A. ligamentina</i>	166	72.3	<i>A. plicata</i> (13.3), <i>E. dilatata</i> (1.2), <i>L. siliquoidea</i> (0.6), <i>L. recta</i> (11.4), <i>Q. pustulosa</i> (1.2)
5	Threeridge, <i>A. plicata</i>	20	70.0	<i>A. ligamentina</i> (5.0), <i>E. dilatata</i> (5.0), <i>E. triquetra</i> (20)
31	Giant Floater, <i>P. grandis</i>	30	70.0	<i>A. ferussacianus</i> (3.3), <i>L. costata</i> (26.7)
11	Northern Riffleshell, <i>E. t. rangiana</i>	47	66.0	<i>A. viridis</i> (4.3), <i>A. plicata</i> (2.1), <i>E. dilatata</i> (14.9), <i>E. triquetra</i> (6.4), <i>F. flava</i> (2.1), <i>L. complanata</i> (2.1), <i>L. compressa</i> (2.1)
23	Black Sandshell, <i>L. recta</i>	50	58.0	<i>A. ligamentina</i> (12.0), <i>A. plicata</i> (2.0), <i>L. cardium</i> (4.0), <i>L. fasciola</i> (6.0), <i>L. siliquoidea</i> (14.0), <i>Q. pustulosa</i> (4.0), <i>V. iris</i> (8.0)
10	Spike, <i>E. dilatata</i>	128	53.1	<i>A. ligamentina</i> (0.8), <i>A. plicata</i> (19.5), <i>E. t. rangiana</i> (17.2), <i>E. triquetra</i> (3.9), <i>L. siliquoidea</i> (1.6), <i>L. complanata</i> (3.1), <i>P. fasciolaris</i> (0.8)
15	Wavyrayed Lampmussel, <i>L. fasciola</i>	222	44.1	<i>A. ligamentina</i> (3.2), <i>A. marginata</i> (0.5), <i>A. plicata</i> (0.9), <i>C. tuberculata</i> (26.6), <i>E. triquetra</i> (0.5), <i>L. cardium</i> (6.3), <i>L. siliquoidea</i> (4.5), <i>L. complanata</i> (0.5), <i>L. recta</i> (12.6), <i>P. alatus</i> (0.5)
12	Snuffbox, <i>E. triquetra</i>	40	35.0	<i>A. ligamentina</i> (2.5), <i>A. plicata</i> (35.0), <i>E. dilatata</i> (7.5), <i>F. flava</i> (20.0)
17	Fatmucket, <i>L. siliquoidea</i>	143	25.9	<i>A. ligamentina</i> (0.7), <i>A. plicata</i> (0.7), <i>C. tuberculata</i> (0.7), <i>E. dilatata</i> (4.9), <i>E. t. rangiana</i> (0.7), <i>E. triquetra</i> (2.1), <i>L. cardium</i> (53.1), <i>L. fasciola</i> (5.6), <i>L. complanata</i> (0.7), <i>L. recta</i> (4.9)
	TOTAL	1869	71.1	

Table 7. Classification Function Coefficients for the Ausable River model

	Species	Classification Function Coefficients			
		x ₁	x ₂	x ₃	b
		Log Height	Log Hinge Length	Log Length	(Constant)
1	Mucket, <i>A. ligamentina</i>	2923.680	609.892	1901.196	-6316.931
2	Elktoe, <i>A. marginata</i>	3152.446	632.388	2075.813	-7353.952
4	Slippershell, <i>A. viridis</i>	2767.980	829.061	2119.021	-6952.157
5	Threeridge, <i>A. plicata</i>	2828.011	698.290	1897.233	-6264.903
6	Cylindrical Papershell, <i>A. ferussacianus</i>	3002.323	800.550	2054.179	-7297.350
7	Purple Wartyback, <i>C. tuberculata</i>	3098.008	627.860	1963.573	-6928.914
10	Spike, <i>E. dilatata</i>	2839.910	720.750	1971.136	-6515.272
11	Northern Riffleshell, <i>E. t. rangiana</i>	2787.114	753.363	2007.021	-6548.287
12	Snuffbox, <i>E. triquetra</i>	2769.287	706.584	1927.663	-6216.353
13	Wabash Pigtoe, <i>F. flava</i>	2714.147	743.518	1866.439	-6028.515
14	Plain Pocketbook, <i>L. cardium</i>	2996.326	605.240	1957.979	-6616.599
15	Wavyrayed Lampmussel, <i>L. fasciola</i>	3048.237	618.591	1947.155	-6745.987
17	Fatmucket, <i>L. siliquoidea</i>	2989.182	628.142	1944.545	-6614.274
18	White Heelsplitter, <i>L. complanata</i>	2972.665	767.221	2019.621	-7059.544

		Classification Function Coefficients			
		x_1	x_2	x_3	b
Species		Log Height	Log Hinge Length	Log Length	(Constant)
19	Creek Heelsplitter, <i>L. compressa</i>	2875.981	797.805	2117.349	-7135.602
20	Flutedshell, <i>L. costata</i>	3134.736	809.951	2036.361	-7611.985
21	Fragile Papershell, <i>L. fragilis</i>	2403.713	454.831	1490.437	-4059.382
23	Black Sandshell, <i>L. recta</i>	2998.305	631.780	1880.203	-6492.272
28	Pink Heelsplitter, <i>P. alatus</i>	3372.867	636.236	1754.926	-7149.084
29	Kidneyshell, <i>P. fasciolaris</i>	2812.228	581.056	1848.847	-5878.965
31	Giant Floater, <i>P. grandis</i>	3082.951	812.689	2080.156	-7596.827
32	Pimpleback, <i>Q. pustulosa</i>	3025.939	559.337	1905.054	-6472.337
35	Creeper, <i>S. undulatus</i>	2881.358	851.316	2172.606	-7418.453
38	Deertoe, <i>T. truncata</i>	2270.102	437.607	1407.893	-3633.468
39	Paper Pondshell, <i>U. imbecillis</i>	2961.259	828.369	2019.304	-7174.077

Table 8. Discriminant Function Analysis (DFA) model including only Grand River species

	Species	n	% correct	Misclassified as (%)
2	Elktoe, <i>A. marginata</i>	109	100	--
28	Pink Heelsplitter, <i>P. alatus</i>	20	100	--
32	Pimpleback, <i>Q. pustulosa</i>	14	100	--
4	Slippershell, <i>A. viridis</i>	7	100	--
6	Cylindrical Papershell, <i>A. ferussacianus</i>	7	100	--
39	Paper Pondshell, <i>U. imbecillis</i>	7	100	--
18	White Heelsplitter, <i>L. complanata</i>	6	100	--
19	Creek Heelsplitter, <i>L. compressa</i>	5	100	--
25	Hickorynut, <i>O. olivaria</i>	3	100	--
27	Round Pigtoe, <i>P. sintoxia</i>	191	97.4	<i>T. parvum</i> (2.6)
36	Lilliput, <i>T. parvum</i>	318	94.7	<i>P. sintoxia</i> (1.6), <i>P. fasciolaris</i> (3.8),
35	Creeper, <i>S. undulatus</i>	207	92.8	<i>A. viridis</i> (0.5), <i>L. compressa</i> (1.4), <i>U. imbecillis</i> (5.3)
33	Mapleleaf, <i>Quadrula quadrula</i>	259	88.8	<i>L. fragilis</i> (10.0), <i>T. donaciformis</i> (0.8), <i>T. truncata</i> (0.4)
20	Flutedshell, <i>L. costata</i>	315	86.0	<i>A. ferussacianus</i> (1.0), <i>P. grandis</i> (13.0)

	Species	n	% correct	Misclassified as (%)
26	Round Hickorynut, <i>O. subrotunda</i>	97	84.5	<i>A. ligamentina</i> (6.2), <i>O. olivaria</i> (1.0), <i>P. fasciolaris</i> (8.2)
24	Threehorn Wartyback, <i>O. reflexa</i>	31	77.4	<i>A. ligamentina</i> (3.2), <i>A. plicata</i> (9.7), <i>E. dilatata</i> (6.5), <i>L. siliquoidea</i> (3.2)
41	Rainbow, <i>V. iris</i>	44	77.3	<i>L. cardium</i> (2.3), <i>L. fasciola</i> (18.2), <i>Q. pustulosa</i> (2.3)
14	Plain Pocketbook, <i>L. cardium</i>	225	74.2	<i>A. ligamentina</i> (1.3), <i>E. dilatata</i> (1.3), <i>L. fasciola</i> (6.7), <i>L. siliquoidea</i> (8.9), <i>L. complanata</i> (0.4), <i>L. recta</i> (3.6), <i>O. reflexa</i> (0.9), <i>O. olivaria</i> (2.2), <i>V. iris</i> (0.4)
21	Fragile Papershell, <i>L. fragilis</i>	38	73.7	<i>Q. quadrula</i> (21.1), <i>T. donaciformis</i> (5.3)
31	Giant Floater, <i>P. grandis</i>	30	70.0	<i>A. ferussacianus</i> (6.7), <i>L. costata</i> (23.3)
29	Kidneyshell, <i>P. fasciolaris</i>	33	66.7	<i>A. ligamentina</i> (12.1), <i>O. subrotunda</i> (12.1), <i>T. parvum</i> (9.1)
10	Spike, <i>E. dilatata</i>	128	66.4	<i>A. plicata</i> (16.4), <i>E. triquetra</i> (3.9), <i>L. cardium</i> (0.8), <i>L. siliquoidea</i> (0.8), <i>L. complanata</i> (7.8), <i>O. reflexa</i> (2.3), <i>O. olivaria</i> (1.6)
15	Wavyrayed Lampmussel, <i>L. fasciola</i>	222	65.3	<i>A. ligamentina</i> (2.3), <i>A. marginata</i> (1.4), <i>A. plicata</i> (0.5), <i>L. cardium</i> (5.9), <i>L. siliquoidea</i> (5.4), <i>L. complanata</i> (0.5), <i>L. recta</i> (9.9), <i>O. reflexa</i> (0.9), <i>O. olivaria</i> (2.3), <i>O. subrotunda</i> (0.5), <i>P. alatus</i> (0.5), <i>V. iris</i> (5.0)
37	Fawnsfoot, <i>T. donaciformis</i>	31	58.1	<i>L. fragilis</i> (3.2), <i>T. truncata</i> (38.7)
13	Wabash Pigtoe, <i>F. flava</i>	9	55.6	<i>E. triquetra</i> (11.1), <i>P. sintoxia</i> (33.3)
5	Threeeridge, <i>A. plicata</i>	20	55.0	<i>E. dilatata</i> (5.0), <i>E. triquetra</i> (25.0), <i>O. reflexa</i> (15.0)
1	Mucket, <i>A. ligamentina</i>	166	54.8	<i>A. plicata</i> (7.8), <i>E. dilatata</i> (0.6), <i>L. siliquoidea</i> (0.6), <i>L. recta</i> (5.4), <i>O. reflexa</i> (10.2), <i>O. olivaria</i> (9.0), <i>O. subrotunda</i> (6.0), <i>Q. pustulosa</i> (4.8), <i>V. iris</i> (0.6)

	Species	n	% correct	Misclassified as (%)
23	Black Sandshell, <i>L. recta</i>	50	42.0	<i>A. ligamentina</i> (10.0), <i>A. plicata</i> (2.0), <i>L. cardium</i> (6.0), <i>L. fasciola</i> (2.0), <i>L. siliquoidea</i> (10.0), <i>O. reflexa</i> (2.0), <i>O. oliviara</i> (18.0), <i>Q. pustulosa</i> (2.0), <i>V. iris</i> (6.0)
12	Snuffbox, <i>E. triquetra</i>	40	40.0	<i>A. ligamentina</i> (2.5), <i>A. plicata</i> (27.5), <i>E. dilatata</i> (7.5), <i>F. flava</i> (12.5), <i>O. reflexa</i> (10.0)
38	Deertoe, <i>T. truncata</i>	8	37.5	<i>T. donaciformis</i> (62.5)
17	Fatmucket, <i>L. siliquoidea</i>	143	25.2	<i>A. ligamentina</i> (0.7), <i>E. dilatata</i> (7.0), <i>E. triquetra</i> (0.7), <i>L. cardium</i> (52.4), <i>L. fasciola</i> (2.1), <i>L. recta</i> (1.4), <i>O. reflexa</i> (7.7), <i>V. iris</i> (2.8)
	TOTAL	2783	77.9	

Table 9. Classification Function Coefficients for the Grand River model

		Classification Function Coefficients			
		x ₁	x ₂	x ₃	b
	Species	Log Height	Log Hinge Length	Log Length	(Constant)
1	Mucket, <i>A. ligamentina</i>	987.540	487.721	2084.473	-4094.868
2	Elktoe, <i>A. marginata</i>	1067.681	502.942	2267.059	-4766.871
4	Slippershell, <i>A. viridis</i>	846.511	706.226	2272.086	-4691.096
5	Threeridge, <i>A. plicata</i>	925.349	574.886	2077.230	-4111.885
6	Cylindrical Papershell, <i>A. ferussacianus</i>	965.210	666.902	2245.566	-4822.400
10	Spike, <i>E. dilatata</i>	918.114	597.386	2144.310	-4301.132
12	Snuffbox, <i>E. triquetra</i>	893.881	586.202	2096.111	-4106.568
13	Wabash Pigtoe, <i>F. flava</i>	867.230	621.709	2040.130	-3993.190
14	Plain Pocketbook, <i>L. cardium</i>	1015.446	481.536	2142.373	-4285.884
15	Wavyrayed Lampmussel, <i>L. fasciola</i>	1037.046	491.352	2141.555	-4357.266
17	Fatmucket, <i>L. siliquioidea</i>	1008.577	502.966	2132.292	-4289.177
18	White Heelsplitter, <i>L. complanata</i>	962.747	636.105	2208.567	-4652.253
19	Creek Heelsplitter, <i>L. compressa</i>	902.277	671.889	2282.095	-4769.587
20	Flutedshell, <i>L. costata</i>	1024.716	669.100	2249.297	-4988.616
21	Fragile Papershell, <i>L. fragilis</i>	829.689	355.348	1646.880	-2604.493
23	Black Sandshell, <i>L. recta</i>	1018.561	504.210	2078.934	-4190.237
24	Threehorn Wartyback, <i>O. reflexa</i>	934.152	544.134	2126.909	-4183.444
25	Hickorynut, <i>O. olivaria</i>	1024.417	500.186	2033.353	-4090.319
26	Round Hickorynut, <i>O. subrotunda</i>	998.743	456.684	2023.593	-3920.234
27	Round Pigtoe, <i>P. sintoxia</i>	831.212	584.765	1983.225	-3706.599
28	Pink Heelsplitter, <i>P. alatus</i>	1199.521	487.655	2023.229	-4480.304
29	Kidneyshell, <i>P. fasciolaris</i>	949.037	464.438	2021.630	-3815.015
31	Giant Floater, <i>P. grandis</i>	996.216	675.250	2279.965	-5008.162

		Classification Function Coefficients			
		x_1	x_2	x_3	b
Species		Log Height	Log Hinge Length	Log Length	(Constant)
32	Pimpleback, <i>Q. pustulosa</i>	1044.388	435.713	2096.433	-4156.617
33	Mapleleaf, <i>Q. quadrula</i>	802.893	344.005	1756.639	-2741.115
35	Creeper, <i>S. undulatus</i>	887.202	723.286	2335.567	-4992.274
36	Lilliput, <i>T. parvum</i>	895.289	510.703	1985.437	-3703.088
37	Fawnsfoot, <i>T. donaciformis</i>	786.541	330.185	1564.260	-2336.419
38	Deertoe, <i>T. truncata</i>	781.766	343.156	1556.429	-2333.189
39	Paper Pondshell, <i>U. imbecillis</i>	944.182	693.930	2213.067	-4753.164
41	Rainbow, <i>V. iris</i>	1060.124	494.699	2096.163	-4313.282

Table 10. Discriminant Function Analysis (DFA) model including only Sydenham River species

Species	N	% correct	Misclassified as (%)
2 Elktoe, <i>A. marginata</i>	109	100	--
28 Pink Heelsplitter, <i>P. alatus</i>	20	100	--
32 Pimpleback, <i>Q. pustulosa</i>	14	100	--
7 Purple Wartyback, <i>C. tuberculata</i>	13	100	--
6 Cylindrical Papershell, <i>A. ferussacianus</i>	7	100	--
4 Slippershell, <i>A. viridis</i>	7	100	--
39 Paper Pondshell, <i>U. imbecillis</i>	7	100	--
18 White Heelsplitter, <i>L. complanata</i>	6	100	--
19 Creek Heelsplitter, <i>L. compressa</i>	5	100	--
34 Salamander Mussel, <i>S. ambigua</i>	6	100	--
27 Round Pigtoe, <i>P. sintoxia</i>	191	97.4	<i>T. parvum</i> (2.6)
36 Lilliput, <i>T. parvum</i>	318	94.7	<i>P. sintoxia</i> (1.6), <i>P. fasciolaris</i> (2.2), <i>V. fabalis</i> (1.6)
35 Creeper, <i>S. undulatus</i>	207	92.8	<i>A. viridis</i> (0.5), <i>L. compressa</i> (1.4), <i>U. imbecillis</i> (5.3)
33 Mapleleaf, <i>Quadrula quadrula</i>	259	88.4	<i>L. fragilis</i> (10.4), <i>T. donaciformis</i> (0.8), <i>T. truncata</i> (0.4)
20 Flutedshell, <i>L. costata</i>	315	86.0	<i>A. ferussacianus</i> (1.0), <i>P. grandis</i> (13.0)

	Species	N	% correct	Misclassified as (%)
26	Round Hickorynut, <i>O. subrotunda</i>	97	84.5	<i>A. ligamentina</i> (7.2), <i>P. fasciolaris</i> (7.2), <i>V. fabalis</i> (1.0)
24	Threehorn Wartyback, <i>O. reflexa</i>	31	77.4	<i>A. ligamentina</i> (3.2), <i>A. plicata</i> (9.7), <i>E. dilatata</i> (6.5), <i>L. siliquoidea</i> (3.2)
41	Rainbow, <i>V. iris</i>	44	77.3	<i>C. tuberculata</i> (2.3), <i>L. cardium</i> (2.3), <i>L. fasciola</i> (15.9), <i>Q. pustulosa</i> (2.3)
40	Rayed Bean, <i>V. fabalis</i>	132	75.8	<i>O. subrotunda</i> (2.3), <i>P. sintoxia</i> (0.8), <i>P. fasciolaris</i> (10.6), <i>T. parvum</i> (10.6)
14	Plain Pocketbook, <i>L. cardium</i>	225	74.2	<i>A. ligamentina</i> (1.8), <i>L. fasciola</i> (7.1), <i>L. siliquoidea</i> (8.4), <i>L. recta</i> (4.9), <i>O. reflexa</i> (0.4), <i>O. subrotunda</i> (0.4), <i>S. ambigua</i> (2.2), <i>V. iris</i> (0.4)
21	Fragile Papershell, <i>L. fragilis</i>	38	73.7	<i>Q. quadrula</i> (21.1), <i>T. donaciformis</i> (5.3)
31	Giant Floater, <i>P. grandis</i>	30	70.0	<i>A. ferussacianus</i> (6.7), <i>L. costata</i> (23.3)
11	Northern Riffleshell, <i>E. t. rangiana</i>	47	66.0	<i>A. viridis</i> (2.1), <i>A. plicata</i> (2.1), <i>E. dilatata</i> (12.8), <i>E. triquetra</i> (2.1), <i>F. flava</i> (2.1), <i>L. complanata</i> (2.1), <i>L. compressa</i> (4.3), <i>O. reflexa</i> (2.1), <i>S. ambigua</i> (4.3)
37	Fawnsfoot, <i>T. donaciformis</i>	31	58.1	<i>L. fragilis</i> (3.2), <i>T. truncata</i> (38.7)
1	Mucket, <i>A. ligamentina</i>	166	56.6	<i>A. plicata</i> (10.8), <i>E. dilatata</i> (0.6), <i>L. siliquoidea</i> (0.6), <i>L. recta</i> (8.4), <i>O. reflexa</i> (9.6), <i>O. subrotunda</i> (6.0), <i>Q. pustulosa</i> (6.6), <i>V. iris</i> (0.6)
13	Wabash Pigtoe, <i>F. flava</i>	9	55.6	<i>E. t. rangiana</i> (11.1), <i>P. sintoxia</i> (33.3),
5	Threeridge, <i>A. plicata</i>	20	55.0	<i>E. dilatata</i> (5.0), <i>E. triquetra</i> (25.0), <i>O. reflexa</i> (15.0)
23	Black Sandshell, <i>L. recta</i>	50	54.0	<i>A. ligamentina</i> (12.0), <i>A. plicata</i> (2.0), <i>L. cardium</i> (6.0), <i>L. fasciola</i> (2.0), <i>L. siliquoidea</i> (10.0), <i>O. reflexa</i> (2.0), <i>O. subrotunda</i> (2.0), <i>Q. pustulosa</i> (2.0), <i>V. iris</i> (8.0)
29	Kidneyshell, <i>P. fasciolaris</i>	33	48.5	<i>A. ligamentina</i> (12.1), <i>O. subrotunda</i> (9.1), <i>T. parvum</i> (9.1), <i>V. fabalis</i> (21.2)

Species	N	% correct	Misclassified as (%)
15 Wavyrayed Lampmussel, <i>L. fasciola</i>	222	45.5	<i>A. ligamentina</i> (2.3), <i>A. marginata</i> (1.4), <i>A. plicata</i> (0.5), <i>C. tuberculata</i> (23.0), <i>L. cardium</i> (5.4), <i>L. siliquoidea</i> (4.1), <i>L. recta</i> (12.2), <i>O. reflexa</i> (0.9), <i>O. subrotunda</i> (0.5), <i>P. alatus</i> (0.5), <i>S. ambigua</i> (0.5), <i>V. iris</i> (3.6)
10 Spike, <i>E. dilatata</i>	128	42.2	<i>A. plicata</i> (17.2), <i>E. t. rangiana</i> (11.7), <i>E. triquetra</i> (3.9), <i>L. cardium</i> (0.8), <i>L. siliquoidea</i> (0.8), <i>L. complanata</i> (0.8), <i>O. reflexa</i> (1.6), <i>P. fasciolaris</i> (0.8), <i>S. ambigua</i> (20.3)
12 Snuffbox, <i>E. triquetra</i>	40	40.0	<i>A. ligamentina</i> (2.5), <i>A. plicata</i> (27.5), <i>E. dilatata</i> (7.5), <i>F. flava</i> (12.5), <i>O. reflexa</i> (10.0)
38 Deertoe, <i>T. truncata</i>	8	37.5	<i>T. donaciformis</i> (62.5)
17 Fatmucket, <i>L. siliquoidea</i>	143	22.4	<i>A. ligamentina</i> (0.7), <i>C. tuberculata</i> (1.4), <i>E. dilatata</i> (2.8), <i>E. triquetra</i> (0.7), <i>L. cardium</i> (53.1), <i>L. fasciola</i> (2.1), <i>L. recta</i> (1.4), <i>O. reflexa</i> (5.6), <i>S. ambigua</i> (7.0), <i>V. iris</i> (2.8)
TOTAL	2978	75.1	

Table 11. Classification Function Coefficients for the Sydenham River model

		Classification Function Coefficients			
		x ₁	x ₂	x ₃	b
	Species	Log Height	Log Hinge Length	Log Length	(Constant)
1	Mucket, <i>A. ligamentina</i>	1023.308	462.913	2059.846	-4084.049
2	Elktoe, <i>A. marginata</i>	1105.678	476.620	2241.015	-4755.012
4	Slippershell, <i>A. viridis</i>	898.491	671.838	2233.443	-4664.210
5	Threeridge, <i>A. plicata</i>	966.042	547.107	2048.213	-4096.357
6	Cylindrical Papershell, <i>A. ferussacianus</i>	1011.919	635.249	2211.776	-4801.222
7	Purple Wartyback, <i>C. tuberculata</i>	1091.736	473.007	2138.817	-4459.765
10	Spike, <i>E. dilatata</i>	961.271	568.170	2113.251	-4283.514
11	Northern Riffleshell, <i>E. t. rangiana</i>	929.378	600.536	2137.634	-4338.072
12	Snuffbox, <i>E. triquetra</i>	936.252	557.541	2065.582	-4089.555
13	Wabash Pigtoe, <i>F. flava</i>	910.543	592.417	2008.655	-3974.798
14	Plain Pocketbook, <i>L. cardium</i>	1051.424	456.571	2117.726	-4275.205
15	Wavyrayed Lampmussel, <i>L. fasciola</i>	1072.812	466.387	2117.165	-4346.675
17	Fatmucket, <i>L. siliquoidea</i>	1045.368	477.462	2106.924	-4277.663
18	White Heelsplitter, <i>L. complanata</i>	1007.551	605.659	2176.341	-4633.037
19	Creek Heelsplitter, <i>L. compressa</i>	951.626	638.957	2245.884	-4745.863
20	Flutedshell, <i>L. costata</i>	1069.992	638.043	2216.935	-4968.816
21	Fragile Papershell, <i>L. fragilis</i>	855.450	337.126	1629.711	-2599.390
23	Black Sandshell, <i>L. recta</i>	1054.027	479.388	2054.669	-4179.502
24	Threehorn Wartyback, <i>O. reflexa</i>	974.438	516.721	2098.317	-4168.725
26	Round Hickorynut, <i>O. subrotunda</i>	1031.737	433.517	2001.291	-3911.415
27	Round Pigtoe, <i>P. sintoxia</i>	872.869	556.675	1952.964	-3689.786
28	Pink Heelsplitter, <i>P. alatus</i>	1228.330	466.015	2005.184	-4474.052
29	Kidneyshell, <i>P. fasciolaris</i>	983.626	440.505	1997.800	-3804.979

		Classification Function Coefficients			
		x_1	x_2	x_3	b
	Species	Log Height	Log Hinge Length	Log Length	(Constant)
31	Giant Floater, <i>P. grandis</i>	1043.132	643.351	2246.145	-4986.849
32	Pimpleback, <i>Q. pustulosa</i>	1076.847	412.823	2074.818	-4148.638
33	Mapleleaf, <i>Quadrula quadrula</i>	831.204	324.543	1737.327	-2734.985
34	Salamander Mussel, <i>S. ambigua</i>	984.317	575.931	2130.067	-4396.142
35	Creeper, <i>S. undulatus</i>	940.072	688.208	2296.380	-4964.509
36	Lilliput, <i>T. parvum</i>	932.378	485.312	1959.266	-3690.661
37	Fawnsfoot, <i>T. donaciformis</i>	810.764	313.056	1548.156	-2331.991
38	Deertoe, <i>T. truncata</i>	806.462	325.712	1539.900	-2328.419
39	Paper Pondshell, <i>U. imbecillis</i>	991.837	661.674	2178.370	-4730.649
40	Rayed Bean, <i>V. fabalis</i>	994.414	418.191	1940.294	-3659.429
41	Rainbow, <i>V. iris</i>	1094.460	470.423	2073.019	-4303.538

Table 12. Discriminant Function Analysis (DFA) model including only Thames River species

	Species	N	% correct	Misclassified as (%)
27	Round Pigtoe, <i>P. sintoxia</i>	191	100	--
2	Elktoe, <i>A. marginata</i>	109	100	--
28	Pink Heelsplitter, <i>P. alatus</i>	20	100	--
32	Pimpleback, <i>Q. pustulosa</i>	14	100	--
7	Purple Wartyback, <i>C. tuberculata</i>	13	100	--
6	Cylindrical Papershell, <i>A. ferussacianus</i>	7	100	--
4	Slippershell Mussel, <i>A. viridis</i>	7	100	--
39	Paper Pondshell, <i>U. imbecillis</i>	7	100	--
18	White Heelsplitter, <i>L. complanata</i>	6	100	--
34	Salamander Mussel, <i>S. ambigua</i>	6	100	--
19	Creek Heelsplitter, <i>L. compressa</i>	5	100	--
25	Hickorynut, <i>O. olivaria</i>	3	100	--
36	Lilliput, <i>T. parvum</i>	318	94.7	<i>P. sintoxia</i> (1.6), <i>P. fasciolaris</i> (2.2), <i>V. fabalis</i> (1.6)
35	Creeper, <i>S. undulatus</i>	207	92.8	<i>A. viridis</i> (0.5), <i>L. compressa</i> (1.4), <i>U. imbecillis</i> (5.3)
33	Mapleleaf, <i>Quadrula quadrula</i>	259	89.2	<i>L. fragilis</i> (9.7), <i>T. donaciformis</i> (0.8), <i>T. truncata</i> (0.4)

	Species	N	% correct	Misclassified as (%)
20	Flutedshell, <i>L. costata</i>	315	86.0	<i>A. ferussacianus</i> (1.0), <i>P. grandis</i> (13.0),
26	Round Hickorynut, <i>O. subrotunda</i>	97	84.5	<i>A. ligamentina</i> (6.2), <i>O. olivaria</i> (1.0), <i>P. fasciolaris</i> (7.2), <i>V. fabalis</i> (1.0),
24	Threehorn Wartyback, <i>O. reflexa</i>	31	77.4	<i>A. ligamentina</i> (3.2), <i>A. plicata</i> (9.7), <i>E. dilatata</i> (6.5), <i>L. siliquoidea</i> (3.2)
41	Rainbow, <i>V. iris</i>	44	77.3	<i>C. tuberculata</i> (2.3), <i>L. cardium</i> (2.3), <i>L. fasciola</i> (15.9), <i>Q. pustulosa</i> (2.3)
40	Rayed Bean, <i>V. fabalis</i>	132	75.8	<i>O. olivaria</i> (0.8), <i>O. subrotunda</i> (1.5), <i>P. sintoxia</i> (0.8), <i>P. fasciolaris</i> (10.6), <i>T. parvum</i> (10.6)
14	Plain Pocketbook, <i>L. cardium</i>	225	74.2	<i>A. ligamentina</i> (1.8), <i>L. fasciola</i> (7.1), <i>L. siliquoidea</i> (8.4), <i>L. recta</i> (3.6), <i>O. reflexa</i> (0.4), <i>O. olivaria</i> (1.8), <i>S. ambigua</i> (2.2), <i>V. iris</i> (0.4)
21	Fragile Papershell, <i>L. fragilis</i>	38	73.7	<i>T. donaciformis</i> (2.6), <i>Q. quadrula</i> (21.1), <i>T. donaciformis</i> (5.3)
31	Giant Floater, <i>P. grandis</i>	30	70.0	<i>A. ferussacianus</i> (6.7), <i>L. costata</i> (23.3)
11	Northern Riffleshell, <i>E. t. rangiana</i>	47	66.0	<i>A. viridis</i> (2.1), <i>A. plicata</i> (2.1), <i>E. dilatata</i> (12.8), <i>E. triquetra</i> (2.1), <i>F. flava</i> (2.1), <i>L. complanata</i> (2.1), <i>L. compressa</i> (4.3), <i>O. reflexa</i> (2.1), <i>S. ambigua</i> (4.3)
37	Fawnsfoot, <i>T. donaciformis</i>	31	58.1	<i>L. fragilis</i> (3.2), <i>T. truncata</i> (38.7)
13	Wabash Pigtoe, <i>F. flava</i>	9	55.6	<i>E. t. rangiana</i> (11.1), <i>P. sintoxia</i> (33.3)
5	Threeridge, <i>A. plicata</i>	20	55.0	<i>E. dilatata</i> (5.0), <i>E. triquetra</i> (25.0), <i>O. reflexa</i> (15.0)
1	Mucket, <i>A. ligamentina</i>	166	53.6	<i>A. plicata</i> (8.4), <i>E. dilatata</i> (0.6), <i>L. siliquoidea</i> (0.6), <i>L. recta</i> (5.4), <i>O. reflexa</i> (9.6), <i>O. olivaria</i> (9.0), <i>O. subrotunda</i> (5.4), <i>Q. pustulosa</i> (6.6), <i>V. iris</i> (0.6)
29	Kidneyshell, <i>P. fasciolaris</i>	33	48.5	<i>A. ligamentina</i> (12.1), <i>O. subrotunda</i> (9.1), <i>T. parvum</i> (9.1), <i>V. fabalis</i> (21.2)

	Species	N	% correct	Misclassified as (%)
15	Wavyrayed Lampmussel, <i>L. fasciola</i>	222	45.5	<i>A. ligamentina</i> (2.3), <i>A. marginata</i> (1.4), <i>A. plicata</i> (0.5), <i>C. tuberculata</i> (23.0), <i>L. cardium</i> (5.4), <i>L. siliquoidea</i> (4.1), <i>L. recta</i> (9.9), <i>O. reflexa</i> (0.9), <i>O. olivaria</i> (2.3), <i>O. subrotunda</i> (0.5), <i>P. alatus</i> (0.5), <i>S. ambigua</i> (0.5), <i>V. iris</i> (3.6)
10	Spike, <i>E. dilatata</i>	128	42.2	<i>A. plicata</i> (16.4), <i>E. t. rangiana</i> (11.7), <i>E. triquetra</i> (3.9), <i>L. cardium</i> (0.8), <i>L. siliquoidea</i> (0.8), <i>L. complanata</i> (0.8), <i>O. reflexa</i> (1.6), <i>O. olivaria</i> (1.6), <i>S. ambigua</i> (20.3)
23	Black Sandshell, <i>L. recta</i>	50	42.0	<i>A. ligamentina</i> (10.0), <i>A. plicata</i> (2.0), <i>L. cardium</i> (6.0), <i>L. fasciola</i> (2.0), <i>L. siliquoidea</i> (10.0), <i>O. reflexa</i> (2.0), <i>O. olivaria</i> (18.0), <i>Q. pustulosa</i> (2.0), <i>V. iris</i> (6.0)
12	Snuffbox, <i>E. triquetra</i>	40	40.0	<i>A. ligamentina</i> (2.5), <i>A. plicata</i> (27.5), <i>E. dilatata</i> (7.5), <i>F. flava</i> (12.5), <i>O. reflexa</i> (10.0)
38	Deertoe, <i>T. truncata</i>	8	37.5	<i>T. donaciformis</i> (62.5)
17	Fatmucket, <i>L. siliquoidea</i>	143	22.4	<i>A. ligamentina</i> (0.7), <i>C. tuberculata</i> (1.4), <i>E. dilatata</i> (2.8), <i>E. triquetra</i> (0.7), <i>L. cardium</i> (53.1), <i>L. fasciola</i> (2.1), <i>L. recta</i> (1.4), <i>O. reflexa</i> (5.6), <i>S. ambigua</i> (7.0), <i>V. iris</i> (2.8)
	TOTAL	2981	74.8	

Table 13. Classification Function Coefficients for the Thames River model

		Classification Function Coefficients			
		x ₁	x ₂	x ₃	b
Species		Log Height	Log Hinge Length	Log Length	(Constant)
1	Mucket, <i>A. ligamentina</i>	1013.092	466.344	2055.717	-4070.546
2	Elktoe, <i>A. marginata</i>	1094.528	480.499	2236.523	-4739.248
4	Slippershell Mussel, <i>A. viridis</i>	890.517	673.602	2228.682	-4650.875
5	Threeridge, <i>A. plicata</i>	956.875	549.759	2044.050	-4083.642
6	Cylindrical Papershell, <i>A. ferussacianus</i>	1002.579	637.703	2207.244	-4786.750
7	Purple Wartyback, <i>C. tuberculata</i>	1080.781	476.742	2134.572	-4444.803
10	Spike, <i>E. dilatata</i>	952.206	570.732	2108.902	-4270.407
11	Northern Riffleshell, <i>E. t. rangiana</i>	920.795	602.785	2133.175	-4325.146
12	Snuffbox, <i>E. triquetra</i>	927.438	560.018	2061.326	-4077.071
13	Wabash Pigtoe, <i>F. flava</i>	902.230	594.512	2004.530	-3962.945
14	Plain Pocketbook, <i>L. cardium</i>	1040.828	460.216	2113.475	-4260.930
15	Wavyrayed Lampmussel, <i>L. fasciola</i>	1062.041	470.061	2112.951	-4332.121
17	Fatmucket, <i>L. siliquoidea</i>	1034.954	480.941	2102.700	-4263.554
18	White Heelsplitter, <i>L. complanata</i>	998.136	608.244	2171.895	-4618.898
19	Creek Heelsplitter, <i>L. compressa</i>	942.912	641.164	2241.162	-4731.880

		Classification Function Coefficients			
		x_1	x_2	x_3	b
Species		Log Height	Log Hinge Length	Log Length	(Constant)
20	Flutedshell, <i>L. costata</i>	1060.050	640.731	2212.480	-4953.562
21	Fragile Papershell, <i>L. fragilis</i>	846.733	340.217	1626.508	-2590.450
23	Black Sandshell, <i>L. recta</i>	1043.577	482.842	2050.607	-4165.639
24	Threehorn Wartyback, <i>O. reflexa</i>	964.981	519.646	2094.017	-4155.584
25	Hickorynut, <i>O. olivaria</i>	1048.147	479.501	2006.199	-4066.733
26	Round Hickorynut, <i>O. subrotunda</i>	1021.331	437.111	1997.333	-3898.180
27	Round Pigtoe, <i>P. sintoxia</i>	864.448	558.914	1948.270	-3676.754
28	Pink Heelsplitter, <i>P. alatus</i>	1216.016	470.246	2001.530	-4458.067
29	Kidneyshell, <i>P. fasciolaris</i>	973.768	443.847	1993.779	-3792.387
31	Giant Floater, <i>P. grandis</i>	1033.475	645.920	2241.566	-4971.727
32	Pimpleback, <i>Q. pustulosa</i>	1065.795	416.808	2070.716	-4134.281
33	Mapleleaf, <i>Quadrula quadrula</i>	825.603	326.217	1733.181	-2728.099
34	Salamander Mussel, <i>S. ambigua</i>	975.034	578.559	2125.708	-4382.635
35	Creeper, <i>S. undulatus</i>	931.689	690.107	2291.510	-4950.226
36	Lilliput, <i>T. parvum</i>	923.326	488.121	1955.287	-3678.946
37	Fawnsfoot, <i>T. donaciformis</i>	802.466	316.029	1545.108	-2323.939

		Classification Function Coefficients			
		x_1	x_2	x_3	b
Species		Log Height	Log Hinge Length	Log Length	(Constant)
38	Deertoe, <i>T. truncata</i>	798.282	328.580	1536.872	-2320.463
39	Paper Pondshell, <i>U. imbecillis</i>	982.875	663.846	2173.909	-4716.624
40	Rayed Bean, <i>V. fabalis</i>	984.377	421.664	1936.447	-3647.063
41	Rainbow, <i>V. iris</i>	1083.521	474.122	2068.965	-4288.999